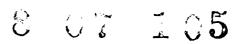
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	biominera	11.					
Both apo and holo ferr: Mn ²⁺) under anaerobic condit protein sites whereas holo t Holo ferritin undergoes reduphase. Apo ferritin also un Electron transfer reactions has relatively high electric	itin bind Fe [†] a tions as a func binds large num uction at its F ndergoes redox occur readily	s well as oth tion of pH. bers of Fe2+ e00H mineral reactions pro in the Fe00H	Apo ferrit on its min core formi esumable at	in bir eral on ng a I some ating	nds 8 1 core su Fe2+ mi amino	re2+ aurface ineral acid ineral	at e. L cite
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ANNUAL REPORT:

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PRINCIPAL INVESTIGATOR:

Gerald D. Watt

CONTRACTOR:

The University of Colorado

CONTRACT TITLE:

Chemical and Biochemical Properties of

the Iron Mineral Core of Mammalian

Ferritin

START DATE: 6/1/88

RESEARCH OBJECTIVES: To evaluate the chemical composition of the iron core of mammalian ferritin with regard to: 1)

3) electron transfer reactions.

PROGRESS (YEAR 1)

Metal Ion Binding. Holo mammalian ferritin contains up to 4500 iron atoms within its hollow protein interior (80A in diameter) in the form of an FeOOH mineral core. Removal of this iron core by reduction and Fe(II) chelation forms apo ferritin with an empty, hollow interior. We have shown that both apo and holo ferritin avidly bind Fe2+ under anaerobic conditions in a pH dependent binding process. For holo ferritin, the binding occurs at the FeOOH mineral surface while for apo ferritin, Fe2+ binds at 8 protein sites. Cu2+. 2n2+ and Mn2+ also strongly bind to both holo and apo ferritin. These metals are readily removed by chelating agents. Immobilization of both ferritin forms on acrylamide or sephanose supports readily occurs with complete retention or of this metal ion binding ability.

pinding of metal ions; 2) electrochemical energy storage and

Redox Chemistry. The mineral core of mammalian ferritin undergoes complete reduction to a corresponding Fe2+ core (of unknown composition and structure at present). The reduction potential is -310 mv at pH 8 but is strongly pH dependent. suggesting the following overall reaction: FeOOH + e + 2H+ = FeOH+ + H_2O . The mineral core of ferritin \tilde{i} is thus capable of storing Targe numbers of low potential electrons as well as protons in its isolated interior. Partial reduction of the ferritin core produces discrete Fe(II) and Fe(III) within the core mineral. الدائلة والأرا

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Electron Transfer. Addition of labeled 57Fe2+ to anaeropic holo ferritin produces 57Fe3+ bound to the mineral core, demonstrating electron transfer to the bulk core and indicating that the mineral core is conductive. Addition of Fe2+ specific chelators removes 57Fe2+, demonstrating the reversibility of this electron transfer reaction. Attempts to determine how redox reagents external to the sequestered mineral core are able to transfer electrons to the interior through the intervening 20-30A protein shell are underway. Three hypotheses are being investigated: 1) Fe2+/Fe3+ mediated electron transfer: 2) electron tunneling and 3) protein mediated electron transfer from internal amino acid residues. Evidence for the latter process has been obtained recently.

WORK PLAN (YEAR 2): Having demonstrated the presence of a reduced (Fe2+) core, attempts to reconvert it into the original Fe00H state will be undertaken. This reactivity will demonstrate whether the redox reactions are reversible. We will also establish whether other metal ions (Zn, Cu etc.) are capable of forming aggregated mineral phases within the ferritin interior. Attempts to form magnetite and other iron aggregates will also be undertaken. Finally, we will investigate in more detail the nature of the electron transfer process through the ferritin protein shell.

INVENTIONS: None

PUBLICATIONS:

1. Redox Reactivity of Bacterial and Mammalian Ferritin: Is Reductant Entry into the Ferritin Interior a Necessary Step for Iron Release?

Proc. Natl. Acad. Sci. USA. (1989) 85, 7457-7461.

2. Redox Reactions Associated with Iron Release from Mammalian Ferritin.

Biochemistry (1989) 28,1650-1655.

3. Fe2+ Binging to Apo and Holo Mammalian Ferritin

In Press. Biochemistry

TRAINING ACTIVITIES: The following undergraduate research students (all US citizens) were supported during the past year: Steve Hilty (Occidental, College): Chris Van Buren (University of Colorado) and Chi Hua Chiu (University of Colorado).